Life Saving Links

LINK TO THIS WEEK

Summer 2025	
General Links DailyScheduleVDSSumm25 this doc Overall Plan & TARGETS (link in this doc) ProtocolLinks_VDSs Student SheetSp25 ARI_VDS_(ARI protocols) ConflictsStudentsVDSSu25 LandingPageSummer25VDS ConflictsStudentsVDSSu25 ResearchPages_VDSs Journal Club papers JournalClubSumm25 ClickUp-VDS (Project management software)	Links to sections below: Week 1, Week 2, Week 3, Week 4, Week 5, Week 6, Week 7 Student Presentations (for Sharebacks) Summ25 Shareback Slides - Summ25 Shareback Slides-Week5Data Archive - (presented week6) - Summ25 Shareback Slides-Week4Data Archive (presented week5) - Summ25 Shareback Slides-Week3Archive (presented week4) - Summ25 Shareback Slides-Week2Archive(presented week3) - Summ25 Shareback Slides-Week2Archive(presented week3) - Summ25 Shareback Slides-Week1Archive(presented week2)
	Targets Target Selection Form - FRI (tbd)
HSRA: HRIWorkFlowCurriculumVirtualCures v.2 HRI Virtual Cures Slide Deck Sign In/Out Sheets:	VR/AR - https://mtzgroup.github.io/molar/ - (use iPhone and view a structure in VR - doesn't work with Android)
Box-HSRAAttendanceUpload (upload pic of attendance sheet at end of each day) HSRA 2025 UT Box upload folder (for VDS images/data/results) - Added Sam to access	AI - Microsoft Copilot (via UT), ChatGPT - AlphaFold Molecular Dynamics

Overall Goal - HSRA (on this doc) A Box folder to submit your site-specific safety training performed with HSRA participants	
DI Training Slides HSRA Participant Poster Template for poster symposium on Wednesday, July 16.	
HSRA general FAQ	
Remind	
OLD LINKS UNIFIEDLabSched VDSSumm23 - Google Sheets DO WE NEED THIS? DailyScheduleVDSFa23 DailyScheduleVDSSumm23	qPCR Machine Schedule SquawkSheet VDSclass Google Drive
	Summer 2024, etc ConflictsStudentsVDSSu24 DailyScheduleVDSSumm24 LandingPageSummer24VDS (with names o' folks)
	ShareBackSlides_VDS_Su22

ProtocolsVDSs ProtocolLinks VDSs KnownDNASequencesVDS

Table o' Contents

Life Saving Links

STAFF NOTES

Research Pages

WEEKLY LINKS and Dates:

WEEK 1 - Monday June 9th - Friday June 13th, 2025

OVERVIEW

HSRA specific

30,000 Foot Schedule Overview

NDM-1 Variants Project

Overall Plan & TARGETS

<u>ARIers</u>

Overall Goal - ARI

Specific Aims - ARI V

Overall Goal - FRI

Specific Aims - FRI -

Overall Goal - HSRA

Specific Aims - HSRA

UPCOMING WEEKS

WEEK 2 - Jun-16 Monday - TO BE UPDATED

WEEK 3 - Jun-23

WEEK 4 - June 30th Monday +++++++++++++++LINK TO TOP

WEEK 5 - Jul-7 Monday ++++++++

WEEK 6 - Jul-14 Monday, 2025

- JC#3 - WEEK 6

WEEK 7 - July-21 Monday, 2025

OLD STUFF BELOW HERE to be updated 4/21/23

Results & Data

THINGS TO DO WHEN EXTRA TIME:

PROTOCOL LINKS for Copying and Pasting

PAST WEEKS

STAFF NOTES

5/12/25 - Sam, Antonia, Dr. B

Pic - post to Insta - DONE

Inventory - Done

organize Lab freezers and fridges - DOne

Organize paper shelf/rack

Clean profiles off desktop computers for the Nanodrops -

Make single sign on - DR. B

Laminator alligator - lab advertisement - DONE

Update this doc with dates, etc

Conflict SCheduel - Antonia

HRI stuff - - Sam

- Look at what we did last year
- View Syvia's slides, copy, share, update

Mentors

- HSRA Orientation - DI Volunteer Sign Up 2025.xlsx

ARI: Sal, Celeste, Megan, Sharon, Ifeany (Research)

FRI: Antonia, Sam, Pooja

Researcher Names:

ARI

Helen, Hannah, Riya, Mai, laura, Haiga, Abiel, Dante, Shaun, Kiana, Shriya, lan

IndRes

Aileen, Haneef, Vinu

FRI

Nik, Keertana, Karthik, Apri, Bedour, Ariel, Skye, Patrick, Abigail, Xiya, Ganesh, Ariana, Rhea, Nicole, Manal

HSRA

Harihar, , Laila, , Dhiya, , Akshitha, Srinidh, Jonathan, Sam, Nicholas

Research Pages

<u>LinksToResearchPages_VDSs</u>

- VDSshared ResearchPage

VDSers, Put your links to Research Pages here:

_ _ .

Nikhil Kasturi - 2025 Nikhil Kasturi Research Page VDS-FRI

Hema (Keertana) Vemuri- ■ 2025_KeertanaVemuri_ResearchPageVDS-FRI Saikrishna (Karthik) Gundlapalli-

2025_KarthikGundlapalli_ResearchPageVDS-FRI

April White- 2025_AprilWhite_ResearchPageVDS-FRI

Bedour Qaisieh - 2025 Bedour Qaisieh Research Page VDS-FRI

Ariel Chanthalangsy- 2025 ArielChanthalangsy ResearchPageVDS-FRI

Skye Yero- 2025 SkyeYero ResearchPageVDS-FRI

Patrick Kim- ■ 2025 PatrickKim ResearchPageVDS-FRI

Abigail Norris- 2025 AbigailNorris ResearchPageVDS-FRI

Xiya Chen- 2025 XiyaChen ResearchPageVDS-FRI

Ganesh Annavajjala = 2025 Ganesh Annavajjala Research Page VDS-FRI

Ariana Thompson- ■ 2025_ArianaThompson_ResearchPageVDS-FRI
Nicole Marin- ■ 2025_NicoleMarin_ResearchPageVDS-FRI
Rhea Ahuja- ■ 2025 RheaAhuja ResearchPageVDS-FRI

2024 dh39566 DoraHalder VirtualScreeningNotebookVDS-ARI

Tip: use the document outline at the left to navigate.

ARI - VDS (to be updated)

- ARI-VDS Summer 2023 Schedule
- ARI-VDS Student Plasmid Archive
- DSF Template ***Make a copy of this and edit that

STANDING MENTOR TASKS:

 Make dilutions of DNA ladders?? - ask Dr. B how - sometime in 1st or 2nd week week

-

GSK-3 beta specific info

Google Folders in VDSclass/Projects: GSK-3beta

Google Doc: GSK-3beta

- <u>50-153-2778</u> glutathione resin.
- AAJ6216606 glutathione
- Plasmid from AddGene: <u>GSK3 beta pGEX Plasmid #15898</u>) <u>Human, has GST tag</u>
- Affinity GST Purification using Glutathione Bulk Resin

Links to weeks: Week 1 Week 2 Week 3 Week 4 Week 5 Week 6 Week 7

WEEKLY LINKS and Dates:

LINK TO THIS WEEK 2025

Week 1 Mon June 9th, 2025

Monday - 1st Group Meeting

Intro, Unboxing!, buy supplies at store, make plates, LB

Transform all viable target plasmids,

Create Research Pages online

Start notebooks (paper)

HSRA starts

Shadow a Scientist - Middle Schoolers (Tues) - NONE this week

Target Selections

Plasmid/Primer Design

Week 2 Mon June 16th, 2025

Sharebacks

JClub #1

Shadow a Scientist - Middle Schoolers - on Tues

Week 3 Mon June 23rd, 2025

Sharebacks

Shadow a Scientist - Middle Schoolers - on Tues

Week 4 Mon June 30th, 2025

Sharebacks

JClub #2

Shadow a Scientist - Middle Schoolers - on Tues Thurs (July 3rd), Friday (July 4th) - HSRA off

Week 5 Mon July 7th, 2025

Sharebacks

Shadow a Scientist - Middle Schoolers - on Tues

Week 6 Mon July 14th, 2025

Sharebacks

Shadow a Scientist - Middle Schoolers - on Tues

HSRA last week to Wednesday (HSRA poster session) - Wed July 16th

JClub #3

Week 7 - Mon July 21st, 2025 Final Sharebacks

End

THIS WEEK	++++++++++++++++++++++++++++++++++++++
+++++++	++++++++++++++++++++++++++++++++++++++

WEEK 7 - July-21 Monday, 2025

OVERALL ITEMS FOR WEEK 7 (by group)

ALL

- Group Meetin and Sharebacks on Monday

ARIers

- Continue protein work
- Continue SDM work on variants

- Enzyme assay trial run on NDM

FRIers

- VPipe and IPipe PCRs for PIPE cloning
- -
- VR Goggles protocol
 - A&B -
 - C&D everyone (using your Target protein)
- Plasmid Stock Depositions
 - 2 tubes for Plasmid Stock (Julio freezer)
 - 1 tube for Dr. B's Plasmid Vault (Gregory freezer)
- Enzyme Assay with EhPTP phosphatase
 - <u>ProtocolEnzymeTest_Phosphatase_VDS_Fall13_varyEnzyme</u>
 - Work in pairs or triples

MENTORS:

_

DAY TO DAY ITEMS

Monday - DATE July 21nd, 2025 ------

ALL

- 1:30 pm (WELCH 2.110) SHAREBACKS
 - Summ25 Shareback Slides

FRIers

- Try enzyme assays with EhPTP a

ARIers

MENTORS:

ENTER WHAT WAS COMPLETED TODAY (so we can plan for tomorrow).....

-

Tuesday—-----July 22nd, 2025------

ALL

- Be sure to deposit your plasmids!
 - 2 in stocks (either FRI Plasmid Stocks or ARI Plasmid Student Archive respectively).
 - 1 for Dr. B's Vault
 - VDS Plasmid Stocks (Students)
 - ARI-VDS Plasmid Student Archive
 - VDS Plasmid Vault (Dr B)

.

FRIers

-

ARIers

- Try to run an Enzyme assay (with Sophie)
- Try to run a protein on FPLC together (with Zach)
 - To see how to do it in general.

MENTORS:

ENTER WHAT WAS COMPLETED TODAY (so we can plan for tomorrow).....

OVERVIEW

Create Research Pages
Choose Targets, make plates, make LB,
Transform, Plasmid archive
Transform, Express, Purify, Characterize, DSF
Variants work

- SDM cloning a new variant
- Express, purify characterize

Clone something new - FRI, HSRA

VR headsets for those that didn't get to it

- Who didn't get to VR in spring?:
 - FRI

- ?

- ARI:??

-

Online labnotebooks/research pages, paper notebooks
Do something fun/social
Sharebacks - Monday
Journal Club

- Journal: Journal Club
 - Where you journal (write) about how you perceive what you re

_

30,000 Foot Schedule Overview

Goal

- Protein Projects: make protein, run DSF assays,
 - ARI
 - FRI

- Cloning Projects: archive and deposit plasmids, clone a new one
 - ARI Variants
 - Use SDM to 'clone'
 - FRI for expediency just use past primers (skip design?)
 - All work on 1 Target first, then next
 - FRI Purification trackers
 - Maybe shift everyone to making Purification Tracker for their Target
 - Design primers for adding GBR22 on
 - Include a TEV site?

- HSRA

Clone TEM-1b+GBR22

Need to design primers

Would this be a colorimetric indicator of expression levels?

_

Try this out - TBD Dr. B

QUBES - Resources: Learning Journal Assignment on Biofilms

LINK TO TOP

NDM-1 Variants Project

NotesNDM-1 variants NDM-1 variants folder NDM-1 Variants Sheet NDM-1 Info Sheet (Fast Lab) NDM-1 DNA Sequence

ProtocolStepsForDeterminingAGoodMutant VDS-ARI HRI v3.3

Link to Overall Plan & Targets (on this page)

_

Overall Plan & TARGETS

ARIers

Overall Goal - ARI -

- Identify novel inhibitors against an infectious disease target protein (Spring target usually)
- Create an variant and assay it's functionality relative to your Target

Specific Aims - ARI

MAIN TARGET

- DNA Work on Target
 - LB agar plates, Transform plasmid into DH5alpha, o/n small culture, mini-prep, DNA Sequencing, Plasmid Stock Deposition
- Express, purify, characterize a target, DSF on Target
 - LB agar plates
 - Transformation of Target plasmid (s) into BL21(De3)
 - Make Autoinduction Media
 - Expression of target protein
 - Sonicator
 - Make Purification Buffers
 - Affinity purification of target
 - Homemade SDS-PAGE
 - Characterization
 - Dry Gels
 - DSF of Lysozyme
 - DSF Kinetic vs Thermodynamic Lysozyme
 - DSF of Target
 - DSF Kinetic vs Thermodynamic Target
 - Protein Storage Comparison by DSF on Target
 - Ion Exchange
 - Crystallization trays apo or substrate bound
 - Order Compounds + Pos Ctrl

- DSF of protein and ligand (x 2)
- enzyme assay on target
- Inhibition assay

_

- Virtual Screening (on Target) (High Priority)
 - Program To Use
 - GOLD & ICM
 - Virtual Screening Protocol Part 1, Part 2, Part 3
 - Docking Strategy
 - select structure
 - select Control Ligands & LigPrep
 - controls docking
 - small libraries docking
 - HF9 & Zinc docking
 - Large Libraries List docking
 - Dock In House Compounds
 - SCORCH
- Virtual Reality on Target
 - Make a session in VR showing a walkthrough of your Target and a positive control, a negative control, and your top hit from screening
- Variant
- Make Variant plasmid & protein
 - Site-Directed Mutagenesis (SDM) to make Variant, o/n small culture, mini-prep, DNA Sequencing, Plasmid Stock Deposition
 - Transform variant into BL21(DE3), express, purify, characterize a target
 - DSF on Variant (compare to parent DSF)
- Virtual Screening on Variant vs Parent
 - Choose ICM or GOLD
 - Virtual Screening Protocols...

FRIers

Overall Goal - FRI

- Identify novel inhibitors against an infectious disease target protein (an existing target from the VDS stable of plasmids) (High Priority)
 - See Targets page

_

- Cloning Projects (will work on One or Two of these)
 - Maybe we all work on 1, then roll to next once successful
- Clone one of the 3 new targets: (maybe just 2 of these)
 - Have GBlocks designed for PIPE cloning:
 - YpDHFR:
 - Pf6PGDH:

_

- Clone GBR22 into EhPTP-pNIC-Bsa4 to make a fusion protein
 - Colorimetric and enzymatic
 - LINK to primer info that Hubert and Quoc made? TBD
 - they got the primers (or designed them) yes has order date of 7/12/2023 in IDT

_

- Clone GBR22 into KPC-2-pNIC-Bsa4 (for long haul truckers) maybes
 - Colorimetric and enzymatic
 - Need to design primers for this

Plasmid Stock Deposition (may not be on your main target)

- YpDalaDala- Yersinia pestis Ariana
- SaDalaDala- Staphylococcus aureus none
- YpFabV Yersinia pestis none
- SaDHFR Staphylococcus aureus none
- PfFabG Plasmodium falciparum *none*/Manal
- pGBR22 *none*
- pNIC-Bsa4 Xiya
- EhPTP none
- FtHap *none*
- MtDalaDala Keertana, Nicole
- EcDHFR Karthik
- Lmaj P. Reductase April
- PfDXR Bedour
- PaDXR Rhea
- YpFabG Ariel
- RpFabG Skye
- MtFabG Patrick
- MtInhA pNIC-Bsa4 Abby
- MtInhA pET-18b Nik
- GSK-3 beta Ganesh

Specific Aims - FRI -

- DNA Work on Target
 - LB agar plates, Transform plasmid into DH5alpha, o/n small culture, mini-prep, DNA Sequencing, Plasmid Stock Deposition -
 - Glycerol Stocks
 - AddGene Deposition on your protein target
- Express, purify, characterize a target, DSF on Target
 - Target Selection Form
 - LB agar plates
 - Transformation of Target plasmid (s) into BL21(De3)
 - Make Autoinduction Media
 - Expression of target protein
 - Sonicator
 - Make Purification Buffers
 - Affinity purification of target
 - Homemade SDS-PAGE
 - Characterization
 - Dry Gels
 - DSF of Lysozyme
 - DSF of Target
 - Protein Storage Comparison by DSF on Target
 - Make glycerol stocks of your BL21(DE3) cells with target plasmid
- Virtual Screening (on Target) (High Priority)
 - Virtual Screening Protocol Part 1, Part 2, Part 3
 - Docking Strategy
 - select structure
 - select Control Ligands & LigPrep
 - GOLD controls docking
 - GOLD small libraries docking
 - GOLD HF9 & Zinc docking
 - GOLD Large Libraries List docking
 - Dock In House Compounds
 - ICM repeat all controls/libraries
 - SCORCH
- Virtual Reality VR (on Target)
 - Make a session in VR showing a walkthrough of your Target and a positive control, a negative control, and your top hit from screening
- PIPE Cloning (of the 5 newer Targets)
 - Primer design Tail and GBlock done in Summer23

- Order tail primers
- GoTo Practice Cloning Sequence -
- Dilute vector primers V-PIPE -
- Vector PCR(VPipe) -
- Dilute GBlock -
- Dilute Tail Primers (iPIPE) -
- Insert PCR (iPIPE) on GBlocks-
- PIPE Cloning -
- Transformation (Dh5alpha)
- Grow o/n colonies (of DH5alpha)
- Plasmid Stock Deposition
- AddGene Deposition
- **GoTo** Protein Sequence
- Transform plasmid into DH5alpha, o/n small culture, mini-prep, DNA Sequencing, store verified plasmid in Plasmid Stocks

HSRA

Overall Goal - HSRA

- HRI Project
 - HRIWorkFlowCurriculumVirtualCures v.2

_

- Make more TEM1-b and EhPTP stocks
 - Plasmid and DNA
 - Make glycerol stocks
- Develop the HRI beta-lactamase plan
 - Test out different targets, IPTG, discs, etc
 - AMp, Tazo, Cef, others
 - Try TEM-1b, EhPTP, KPC-2, NDM-1 (maybe),
 - Try ecDHFR with trimethoprim
 - Optimize antibiotic and IPTG amounts
 - Try autoinduction on discs? for o/n induction vs IPTG
 - DSF of TEM1-b

ALpha Fold

- GBR22 alpha fold structure in order to visualize in PyMOL
- Clone a new target YpFabG
- Virtual Reality of TEM-1
- 3D printing TEM-1b

- https://3d.nih.gov/users/mariusmihasan
- s guideline for lowâ•'cost 3D printing of macromolecules usable for teaching and demonstration
- https://3d.nih.gov/entries/3DPX-007355

-

- Or maybe FtFabl

HSRA specific

- P&A,
- HRI Project
- iCn3D protocol
- AlphaFold ?
- VR Headsets on TEM1b
- Buffer Titration, Beer's law? skip for now
- PyMOL skip for now
- Clone target PIPE cloning skip for now
- Clone EhPTP-pGBR22 ? skip for now
- Protein E, P, C on TEM1b yes
- Protein E, P, C on EhPTP yes
- Protein E, P, C on KPC-2 maybe

-

- Reference:
- Dr. Govea's results
 - VDR: Virtual Cures Results
 - ^ use this as a good sneak peak into good plate labeling and drug concentration strategies
- Purification Trackers

```
- Clone make GBR22 + Target
- Primary: EhPTP+GBR22
- Design of EhPTP + GBR22 plasmid
- For PIPE cloning
- EhPTP + pGBR22
- Secondary:
- TEM1b+GBR22
- KPC-2+GBR22
```

Clone YpDHFR - do we have this one already (Joanna?) -

_

Specific Aims - HSRA

<u>-</u> ./- -1

Week 1 Mon June 9th, 2025

Week 2 Mon June 16th, 2025

Antibiotic Disc assays Protein Expression Glycerol Stocks

Week 3 Mon June 23rd, 2025

Make Tazo discs

Antibiotic Disc assays - Sam to decide on order of how to do these

- Can we make a Matrix for this.
- Add in IPTG
- Add Amp (Hi, med, lo)
- Add in Tazobactam (Hi, med, Io)
- Add in Cefdinir (Hi, med, lo)
- And then combinations (will need to pick a concentration)
- Then varying concentrations of Antibiotics

Protein Purification
Protein Characterization

Matrix - Antibiotic Testing

	IPGT (L, M, H)	AMP (L, M, H)	Tazo (L, M, H)	Cef (L, M, H)	Result?
TEM1b	-	L			
TEM1b	L				

TEM1b	-		
TEM1b	L		
EhPTP			

Week 4 Mon June 30th, 2025 ANtibiotic Disc assays

Protein DSF?

Thurs (July 3rd), Friday (July 4th) - HSRA off

iCN3d - HRI protocols VR Goggles - HRI protocol

Week 5 Mon July 7th, 2025

Generate and gather all final data Make poster

ALphaFold of TEM1 - need Google account?

Alpha Fold of GBR22 - (not related to project)

- Get sequence from Known DNA Sequences
- Send ALphaFold models to Dr. B
 - Dr. B to create an HSRA 2025 UT Box upload folder
 - HSRA 2025 UT Box upload folder

Week 6 Mon July 14th, 2025
Lab group meeting - practice presenting
Field trip?
HSRA last week to Wednesday (HSRA poster session) - Wed July 16th

++++++++++++++++++++++++++++++++++++++
++++++++++++++++++++++++++++++++++++++
LINK TO TOP
UPCOMING WEEKS
++++++++++++++++++++++++++++++++++++++
++++++++++++++++++++++++++++++++++++++

OLD STUFF BELOW HERE _ to be updated 4/21/23

Results & Data

Upload shared files to these folders. Post 'highlights' for each protocol or procedure that you do. Add direct links here once made on the Research Pages page of VDS Portal.

Research Pages - on VDS Portal VDSshared ResearchPage - possibly remote work

2022 ARI - ARI Student-Researchers Folders

• Research Pages: <u>Anish Holden Hafsah Jose Griffin Hriman Varsha</u> <u>Shreyaa Humberto Lorena</u>

2022 FRI - FRI Student-Researchers

- Folders: Rachel Rida Quoc Valeria Hubert Sylvia Mia
 - o <u>Krista</u> <u>Naturi</u>
- - Su2022_hj7654_HubertJaegy_ResearchPageVDS
 - Su2022 mlt3375 MiaTernus ResearchPageVDS
 - O VDS Club: ??

HSRA Folks: ???????? Miscellaneous

-OLD: Target Teams (FRI 2020) Notes PaDXP - Google Docs Notes FtFBA - Google

Docs Notes InhA - Google Docs Notes VcUPh - Google Docs Notes SpUGD -

Google Docs

Instructions for Updates to online Research Page: Protocols

- Save a google docs copy of the protocol, modify the protocol according to what you do, and then save a pdf of that protocol to put into student folder or link on the research page for the day
 - Alternatively: Download the protocol as a pdf and save into student research folder and link the PDF onto your own research page
- *YEAR_MONTH_DATE_PROTOCOLNAME* name your pdf protocols according to this format

THINGS TO DO WHEN EXTRA TIME:

Go to BioScience Storeroom (stockroom) to buy stuff

 Give receipt to Dr. B or place in the black receipts basket (can place in box outside office also)

After we get sharpies - Claim your sharpie!

 Write your name on your sharpie & make it look unique with some tape colors/ patterning.

Unboxing

Clean glassware

Clean up the sinks

Empty biohazard buckets

Make more Nanopure water

Make 70% EtoH spray bottle

Make Alconox spray bottle

Make more plates as needed



ProtocolLaminatorAlligatorVDS

 VDSers each laminate a piece of paper with 'whiteboard - VDS' written on top for use as a writing surface for calculations, etc in lab. Place on benchtops

Clarification on 'Output' documents for Summer 2022

FRI and ARI

- 1. Research Page- Student
 - PURPOSE: This is for you to have a centralized document to keep track of your work that is a running log of your work
 - in chronological or reverse chronological order, can have data and analysis in it or
 - Exists within your Student Folder where you can store your files and copies of protocols
 - Granular protocol steps should <u>not be</u> in this document but in a copy of the protocol that you put in your folder.
 - NOTE: if you are on a specific project with someone else you might share a Labnotebook-Project in a separate folder. Just reference to this from your Labnotebook-Student
- 2. Individual student <u>ResearchPagesVDS_UT Google Drive</u>: (hybrid/wet lab students only for now)
 - o PURPOSE: This is for reporting back progress to Dr. B and mentors
 - o To post each of your results of for the week with brief analysis
- 3. ShareBackSlides VDS Su22
 - PURPOSE: 1-2 highlights for the week to share with all of VDS (this replaces the <u>VDS Shared Research Page</u> for this summer)

PROTOCOL LINKS for Copying and Pasting

- Document: ProtocolLinks VDSs

- Folder: ProtocolsVDS

++++++++++++++++++++++++++++++++++++++
++++++++++++++++++++++++++++++++++++++
++++++++++++++++++++++++++++++++++++++
++++++++++++++++++++++++++++++++++++++
PAST WEEKS
++++++++++++++++++++++++++++++++++++++
WEEK 1 - Monday June 9th - Friday June 13th, 2025
OVERALL ITEMS FOR WEEK (by group)
OVERALL HEIMS FOR WILLIN (Dy Gloup)

ALL

- 1st Meeting of Summer @ 2pm on Monday June 9th
- Mentors arrive by 1pm. On Monday

ARIers

- Use Spring Spreadsheet?

FRIers

- Start on AddGene depositions tasks
 - Take over from Spring work, start new one.

HSRA

- Ideal Logic HSRA attendance (Dr. B or Sam to add them)
- Note: no online lab safety for HSRA (only on-site training)
- GUIDELINES for HSRA
 - Note, 2 DI's present at all times with HSRA, Remind App can be used at anytime

- Email is only 8am-6pm M-F
- No SLACK, no GroupMe, no Social Media
- HSRA students can have their own communication group on these type of methods, but DI's can be a part of this
- Sign In & Sign out but once you sign out for day, you cannot return to lab.
 - So no trips away from lab unless with 2 DIs (e.g. grabbing a coffee)
 - 5 min restroom breaks can be solo
- Wed afternoon talks each week
 - Need 2 DI escort to get there.
- No Google Docs
 - So must print protocols
 - UT Box is ok, Canvas is OK



DAY TO DAY ITEMS

Monday - DATE June 9th, 2025 - link to TOP

ALL

- 1st Meeting 2pm in WCH 1.110
- Welcome!
- Update Daily Conflict schedules on the Conflicts spreadsheet
 - Choose day for weekly group meeting probably Mondays
- Update weekly/monthly conflicts on Landing Page as a comment
- Discuss overall plan
- Discuss documents/ navigation
- Discuss HSRA, Shadow A Scientist
- Group selects a weekly meeting time/J club/Sharebacks
- Create Research Pages online DONE for FRI by Antonia
- Start notebooks (paper)
- Mentors
 - Mentors plan mentor shifts for coverage put on sheet
 - ClickUp discuss

FRIers

- _
- _
- -

- Prepare stuff for autoclave for tomorrow
 - Make mixture for plates for FRI
 - Make LB
- Purchasing supplies
 - See the 2nd tab of this spreadsheet for what to get: <u>ConflictsStudentsVDSSu25</u> (spreadsheet)
 - See <u>PurchasingProtocol VDS</u> for how to buy stuff
- Unboxing
- Pickup primers in FNT DONE
- Dilute pLic for and pLic rev primers? DONE
- Dilute Vpipe for and VPipe rev primers? DONE
 - Primer dilution protocol
 - Each person takes one to dilute
 - PrimerDilution (spreadsheet for calculating)
- Random walk? to BioSci Storeroom, NHB Autoclave facilities
 - NHB autoclave training (?)
- Plan for transformations find plasmids

_

HSRA

- Welcome!
- Ideal Logic HSRA attendance (Dr. B or Sam to add them)
- Lab safety walk through
 - Have Manal join?
- Lab safety training online
- Buy lab notebooks in bioscience storeroom mentor purchases but all go there
- MENTORS: after site-specific OH102 lab safety training. You can upload the site-specific training record to the Box link on DI Canvas.
 - Need to print this paper document and have HSRA researchers sign it after training

ARIers

- Prepare stuff for plates and for LB and Autinduction media
- _
- Make plates LINK TO Protocol
- Make LB/ Autoinduction broth
- Plan for practice PCR
- Plan for transformations find plasmids

MENTORS:

ENTER WHAT WAS COMPLETED TODAY (so we can plan for tomorrow).....

- FRI: made primer dilutions, unboxed
- HSRA: Lab safety walkthrough, purchased lab notebooks, unboxed
- ARI: Made LB broth, Agar plates, Base broth

Tuesday - June 10th, 2025

- NOT TIL 2nd WEEK: 2-3:50 - Middle schoolers visit - Mentors in Charge: (changed to Tuesdays this year from Wednesdays)

ALL

- NOTE, for safety compliance, we need to wear long pants even at the front table in lab where we do 'office' stuff.
- Start reading Journal Club article for next week's journal club:
 - Zolli_JuranDHFRbacteriaHTSBioorgMedChemLett2003.pdf

FRIers

- Paste protocols into lab notebook
- Make LB agar & LB
 - Make LB 100 ml per person (can combine in pairs)
- Autoclave plates and LB , once out of autoclave, pour plates
- Lab safety walk through Manal (Mahdia to lead)
- -
- Clean out front fridge to make room for stuff
- Clean out Julio DONE Sam & Antonia
- Make more liquid stock antibiotics (Amp + Kan)
 - Each person makes 10 ml of stock
 - 2 folks for AMP
 - Others make Kan
- Make 10 mm TE *tris actually
- Figure out how to Make alconox
- Figure out how to make bleach in sink bucket
- Figure out how to make 70% etoh in spray bottles
- Figure out how we make labels (good ones for more permanence)

-

Save for Later:

- NHB autoclave training (?) wait
- Discuss Targets with Dr. B
 - Fill out Target Selection form

HSRA

- Do OH102 checklist w/ mentors and sign document (see Monday for info)
- Online Lab Safety training
- Precision & Accuracy lab ?? maybe skip for sake of time
 - Or do while plates cooling
- Make LB agar plates (HRI way?) see HRI
 - HRIWorkFlowCurriculum v.2
- Make parent plates from glycerol stock (or wait til Wed if plates not ready?)

-- Start testing HRI project

ARIers

- Autoclave LB agar plates and LB media
- Make Autoinduction media -
- Test growths of BL21(DE3) plates from spring?
- Test growths of DH5alpha as well
- Wait on these
- Transform NDM-1 parent plasmid into DH5a
 - Do the truncated version, not the lipidated version
 - For plasmid stock archive
 - For NDM variants
- Transform targets into BL21(DE3)

- Transform targets into DH5alpha - if you didn't get to complete plasmid stock deposition in Spring (probably just do this anyways)

- (Dh5alpha)
 - New Version:
- Pick up primers in MBB coordinate with FRi mentors if they already did it
 - Leave some primers in FRI lab for them to dilute
- Primer dilution
 - ProtocolPrimerDilutionVDS
 - PrimerDilution (spreadsheet for calculating)
- Run P-lic PCR ☑ ProtocolPCR pLICsequencing15.xlsx

MENTORS:

ENTER WHAT WAS COMPLETED TODAY (so we can plan for tomorrow).....

- FRI: made kan and amp liquid stock, autoclaved LB media, made LB Agar plates (kan and amp), made 10mm Tris+H2O (stored in Julio door on bottom)
- HSRA: made LB agar+Kan plates, made parent plates using glycerol stocks
- ARI: make plates, dilute primers, test old plates

Wednesday - June 11th, 2025 - link to TOP

Nicole M - to do lab safety walk through training

ALL

- Check out Haily Vu's paper on NDM-1

- Past VDS researcher
- Characterization of Lipidated New Delhi Metallo-β-lactamase Using Synthetic Nanodiscs | Biochemistry

_

FRIers

- Plasmid Hunting (finding plasmids in lab)
 - ProtocolLinks VDSs
 - 3 Plasmid Storage Locations:
 - VDS Plasmid Archive (mentors) VDS Plasmid Stocks (Students) VDS Plasmid
 Vault (Dr B)
 - Also you are empowered to search in past 'personal' boxes of researchers to find plasmids

_

- Transform plates for plasmid prep (DH5alpha) today
 - Transform plates for projects (BL21(DE3)) wait til Thursday
- SHARE the tube of comp cells you only need $\frac{1}{2}$
- Use 1 plate?
- Determine Antibiotic resistance

- Plasmid tube MUST go back to where you found it (unless Dr. B or Antonia gives different guidance)
 - **People:** Nik, Keertana, Karthik, Apri, Bedour, Ariel, Skye, Patrick, Abigail, Xiya, Ganesh, Ariana, Rhea, Nicole, Manal
 - Unassigned:,,

Which Targets? - update this with whether they worked..... *DH5a *BL21

LIGASES:

- D-alanine-D-alanine ligase (YpDalaDala), Yersinia pestis Ariana FEW, how about BL21 ???? (if not BL21- then skip) failed
- D-alanine-D-alanine ligase (SaDalaDala) Staphylococcus aureus -
- MtDalaDala Mycobacterium tuberculosis Keertana, Nicole NONE will need to retransform DH5alpha, second plate didnt grow FEW REDUCTASES
- Dihydrofolate reductase (SaDHFR)- Staphylococcus aureus -
- EcDHFRKarthik, FEW GOOD
- Lmaj P. ReductaseApril, SOME GOOD
- PfDXRBedour, NONE will need to retransform second one grew a few - needs BL21 plate
 - retry DH5alpha in Week 2
- PaDXR ?? Rhea SOME SOME
- YpFabGAriel, FEW FEW
- RpFabGSkye, NONE, #2, none NONE
 - We have some other tubes of this plasmid that we can try DH5alpha agins in Week 2 (dr. B - 6/13/25)
- Pf FabG truncated version Manal
- MtFabG (aka MabA)Patrick, ONE GOOD
- InhA (MtInhA) pNIC-Bsa4 Mycobacterium tuberculosisAbigail,
 ONE FEW
 - - laura in ARI has this also,
- MtlnhA pET-18bNik, FEW GOOD

KINASE

- GSK-3 beta ? non, infectious Ganesh, NONE NONE, #2 GOOD
 - I think we should have a plate we can 'steal' a colony from in
 ARI lab for this. Ask Ifeanyi . Dr. B 6/13/25
 - NOTE this is already in AddGene

PHOSPHATASES

- Transform pGBR22 in DH5 alpha
 - For plasmid prep
 - Maybe for pGBR22 + EhPTP cloning project
 - For Grow up pGBR22 for RE digest
- Transform EhPTP into DH5a HSRA will do
 - For plasmid stock archive
- Transform YopH into DH5a -
 - For plasmid stock archive

_

BACKBONE

- Transform pNIC Bsa4 into dh5a Xiya, Nicole, NONE- needs to retransform RETRANSFORMED DH5a WORKED NONE
 - NOTE this is already in AddGene
 - For plasmid stock archive
 - For practice PCR -will do VPipe as our 'practice' this year
 - For cloning (PIPE cloning)

_

- Make more LB Agar plates wait on this depending if we need them.
- Prepare autoinduction media (<u>at least</u> 1 liter per person)??? NO wait on this this Thursday?
- Prepare sugar mx

_

Learn how to make colliroller prep tubes (6 per microcentrifuge tube)

- V-Pipe PCR
 - pLIC-PCR see
- Target Selection Form maybe tomorrow
 - Please take a look at this form, cogitate on it, and make your selection.
 - We can talk about any of them and answer questions.
 - Let's shoot to submit by Thursday end of day.
 - TargetSelectionFormVDSSumm25

- Enter information into your notebook especially specifics on the plasmid you used
- Copy over Overall Goals and Specific aims to Research Pages (copy from this Daily Schedule)

HSRA

- Streak Plate protocol on homemade plates
- ProtocolTransformationBL21 DE3 VDS v1
- Transform fresh plates and BL21(DE3) [save into DH5alpha for Thursday]
 - Use autoclaved plates that have already been made (have in Grudge Monkey fridge)
 - TEM1b (not TEM1)
 - EhPTP
 - KPC-2 yes
 - Next Steps:
 - Grow overnight cultures for these procedures:
 - Plasmid stock depositions for each target
 - Make glycerol stocks of DH5alpha cells for each target
 - Make glycerol stocks of BL21(DE3) cells for each target

ARIers

- Primer dilution ProtocolPrimerDilutionVDS
- Run P-lic PCR ☑ ProtocolPCR_pLICsequencing15.xlsx
 - Steal pLic for and pLic rev from FRI if you don't have 3
- Make target small cultures 1
- Make overnight cultures for miniprep (target and NDM-1), and for glycerol stock (target) do this closer to end of day 4
- Make sterile 80% glycerol for cell stocks- done
- Make 10X TAE buffer (1 Liter)- have enough
- Resuspended and Dilute some Primers-done
- Make fresh purification buffers-have enough
- Make fresh Tris-HCl buffers (for SDS-PAGE gels)- have enough

MENTORS:

ENTER WHAT WAS COMPLETED TODAY (so we can plan for tomorrow).....

- FRI: transformed plasmids into DH5alpha, added overall objectives and specific aims to research pages, autoclaved 1L base broth, made new LB Agar + Kan plates, took Tris from yesterday and made pH 8
- HSRA: Spreading plates from parent plates (EhPTP, Tem-1b), transformed BL21 for EhPTP, Tem-1b, KPC-2
 - Did a field trip to maker space

 ARI: small cultures, pLIC PCR, overnight culture for miniprep, finished testing fridge plates

Thursday - link to <u>TOP</u>

ALL

- Prepare your slide for Shareback Slides □ Summ25 Shareback Slides □ Shareback Slides □ Summ25 Shareback Slides □ Summ25 Shareback Slides □ Summ25 Shareback Slides □ Shareback Sl
 - Everyone gets a slide make it yours!
- Update your ResearchPage

FRIers

- Transform targets into BL21(DE3)
- Transform EHPTP into BL21(DE3) -
 - For expression for enzyme assay testing
- Transform YopH into BL21(DE3) -
 - For expression for enzyme assay testing

-

- @ EOD (end of day)
- Start overnight cultures for miniprep (do this closer to end of the day)

_

- Other Things to think about (maybe for tomorrow or next week)
 - Homemade SDS- PAGE gels practice
 - PCR VPipe -

-

- Target Disco
 - Talk about targets complete form
- VR goggles for those that haven't completed it
 - See top of document

HSRA

- Jonathan will be out today
- Dhiya out today too
- Check plates
 - RESULT: red- do transformations
 - Do ½ of folkson BL21
 - 1/2 of folks on DH5alpha
- Make some LB

- Transform DH5alpha Tem1b, EhPTP
- Start glycerol stocks save this
- Calc for antibiotic disc making
- Go over powerpoint with them

-

Email Dhiya about fridays in general

_

ARIers

- Mini-prep Target DH5a no growth, need to transform
- Submit to DNA sequencing will complete after successful transformation
- Gel check of PCR- 1/4 worked
- Make target large cultures will have to transform BL21 for most targets

MENTORS:

ENTER WHAT WAS COMPLETED TODAY (so we can plan for tomorrow).....

- FRI: transformed plasmids from Wed 6/11 into BL21, made more LB Agar + Kan plates, purchased BL21 cells, ziplocs, clear packing tape, weigh boats, autoclaved base broth, made 1L sugar mix, started research pages
- HSRA:
- ARI: gel check of PCR, transformed targets into DH5a

Friday - link to TOP

ALL

@ 1pm Move stuff to fridge

_

- ? 1:30 pm ~ 4:00 pm Analyze DNA sequence exercise in the Computer Lab WEL 2.128
 - ProtocolAnalyzeDNASequenceExerciseVDS_v9 (w/ sample sequence)
 - PurpleProteinCodingSequenceforBB.txt

-

Continue wet lab stuff

-

- Prepare your slide for <u>Sharebacks</u> □ Summ25 Shareback Slides
 - Everyone gets a slide make it yours!
- Update your ResearchPage
- Read Journal Club paper over the weekend for Journal Club on ?? of next week
 - https://sites.google.com/site/vdsstream/virtual-drug-screening-stream-a-k-a-virtual-cures/journalclub?authuser=0
 - DHFR paper

FRIers

- Pooja create semesterly boxes for Glycerol Stocks (talk w/ Dr.B)
 - Dh5alpha one and BL21(DE3) one
 - Labels for front of door on -80deg Freezer

_

- ? 1:30 pm ~ 4:00 pm Analyze DNA sequence exercise in the Computer Lab WEL 2.128
 - <u>ProtocolAnalyzeDNASequenceExerciseVDS v9</u> (w/ sample sequence)
 - PurpleProteinCodingSequenceforBB.txt
- Lab notebook check

_

POST a piece of data to Sharebacks slide deck

-

HSRA

- Analyze DNA Sequence Exercise?
 - <u>ProtocolAnalyzeDNASequenceExerciseVDS_v9</u> (w/ sample sequence)
 - PurpleProteinCodingSequenceforBB.txt

-

- OPTIONS
 - iCn3d (do before VR headsets)
 - Day 2 of glycerol stocks
 - Practice making a SDS-Page gel
 - Mini-prep?
- POST a piece of data to Sharebacks slide deck
 - Email your data to Dr. B to post on the slides (copy Sam on the email)

ARIers

- Check plates
- Transform if not done so or if failed (but need mentor to come in on Saturday to move plates) - check with Samtonia

_

- Analyze DNA Sequence Exercise ?
 - <u>ProtocolAnalyzeDNASequenceExerciseVDS v9</u> (w/ sample sequence)
 - PurpleProteinCodingSequenceforBB.txt
- Lab notebook check

- Spin down large culture and put cell pellet in freezer (will continue later).
- DNA workshop thing with Dr. B
- POST a piece of data to □ Summ24 Shareback Slides

MENTORS:

- Lab notebook checks
- Will TACC let us do tours? (TExas Advanced Computing Center at Pickle)
 - https://www.tacc.utexas.edu/about/tours/

ENTER WHAT WAS COMPLETED TODAY (so we can plan for tomorrow).....

- FRI: did Analyze DNA Sequence Exercise, observed growth of BL21 plates, spun down miniprep cultures and stored pellets in Julio, updated research pages
- HSRA:
- ARI:

Saturday

Sunday - link to TOP

DELIVERABLES BY END OF WEEK XXXXX

ALL

- Upload files to: your Student Folder on Google drive or to your Project Folder
- Document your workflow in your paper Labnotebook
- Upload highlights to: ResearchPages
 - This should be several of your 'results' for the week (images, graphs, data)
- Prepare materials for Sharebacks next
 - This should be 1-2 slides of your highlight for the week

WEEK 2 - Jun-16 Monday -

- NOTE: off Juneteenth (19th, Thurs)
- NOTE: no HSRA on Thurs June 19th, 2024

OVERALL ITEMS FOR WEEK (by group)

ALL

- Update Daily Conflict schedules on the Conflicts spreadsheet
- Update weekly/monthly conflicts on Landing Page as a comment

- Journal Club & Sharebacks

_

VR goggles for those that haven't completed it

_

ARIers

- Express protein
 - Someone to express a phosphatase for group (for enzyme assay)
 - Need a bl21 of this get colony from HSRA
- Plasmid Stock Depositions & AddGene Deposition work
- Make homemade SDS-Page gels

FRIers

- Miniprep, plasmid stock deposition & AddGene work
- Glycerol stocks
 - Wait til after positive sequencing
- Express proteins
 - Someone to express a phosphatase for group (for enzyme assay)
 - Need bl21 of this get colony from HSRA
- Pick protein targets
- VPipe PCR
- Make homemade SDS-Page gels (later in week or in Week 3)

HSRA

- Make antibiotic disc
- Antibiotic testing no IPTG
- Glycerol stocks
- Plasmid stock depositions
 - Mini prep, sequencing

MENTORS:

_

DAY TO DAY ITEMS

Monday - DATE June 16th

ALL

- 1 pm (NMS 1.120) WELCH 2.110 SHAREBACKS & Journal Club
 - Summ25 Shareback Slides
 - Note: these may adjust day from week to week
- for Journal Club
 - https://sites.google.com/site/vdsstream/virtual-drug-screening-stream-a-k-a-virtual-cures/journalclub?authuser=0
 - DHFR paper

- Discuss Juneteenth plan (thursday) for Wed
- GBlock design and ordering FRI & HSRA ?

FRIers

- https://docs.google.com/forms/d/e/1FAIpQLSd-hWqCM-Qeuis1z1g6hBbnbNEzhr oVCITWDZEe6AHKY3 EhQ/viewform?usp=header
- ^Target Selection Form
- Transform Tem1b for HSRA in dh5alpha and BL21 DE3 (NICOLE, ABBY)
- Start overnight cutlers of protein targets

. _

Targets determination - Samtonia to update and send Form

-

Review VPipe protocol in advance of tomorrow or next day

-

- Start overnight cultures for plasmids DH5alpha (for the ones that we didn't get to last week)
 - Perhaps everyone just grow up 4 extra tubes of their DH5alpha plates to have back up for plasmid prep
- Start overnight small culture for protein targets
 - ProtocolAutoinductionARI VDS
- Mini-prep, Nanodrop, Submit to DNA Sequencing (save for tomorrow?)
- QIAprep® Spin Miniprep Kit
- Plasmid Stock Depositions
 - <u>PlasmidStockDeposition_Protocol_VDS-FRI</u>
 - QIAprep® Spin Miniprep Kit
 - Nanodrop <u>ProtocolNanoDropVDSa</u>
 - Submit to DNA Sequencing <u>Sequencing ACGT ProtocoVDS</u>

HSRA

- Make antibiliotics
 - AMP and Tazo is fine (do Cefdinier later)
- Make antibiotic discs?
- Start an o/n culture of TEM1b
- make new LB agar plates

-

_

- Make a parent plate of KPC-2 from BL21(DE3) glycerol stock streaking
 - Making a Parent Plate Protocol HRI
 - Used a past plate for KPC-2 instead of glycerol stock

_

- Make Auto induction media -
 - 250 ml per person
 - Use this protocol for making Auto induction (but not for doing the actual expression)
 - ProtocolAutoinductionARI VDS
- Start overnight small culture for protein targets (TEM-1b & KPC-2 & EhPTP)
 - <u>LabProteinExpressionAutoVDS-FRI_v16</u> (FRI protocol from Spring)
 - Scale to 250 ml per person for large culture (vs 25 ml)
 - ?? NOte: if we start this late (e.g on Tues) then on Thursday a mentor needs to move your samples to the fridge

-

ARIers

- Protein work
- Start another round of protein expression
- Start picking NDM-1 variants and find/order primers?
 - <u>Protocol PrimerOrderingInstructionsVDS v2b</u>
 - Site Directed Mutagenesis (SDM) folder
 - SiteDirectedMutagenesisRegularPCR Protocol VDSv1
 - NDM-1 Variants Sheet NDM-1 Info Sheet (Fast Lab) NDM-1 DNA
 Sequence

-

MENTORS:

_

- Update Mentor Schedule
 - Indicate which weeks you will be gone
 - Collectively decide on who will cover which days each week

-

ENTER WHAT WAS COMPLETED TODAY (so we can plan for tomorrow).. -

ARI: additional DH5a transformations, overnight culture, small cultures

FRI: made small cultures for PaDXR, YbFABG, Lmaj P. Reductase, Pet-MtInhA, MtDalaDala, YpDalaDala, MtInhA (pNIC), GSK3 Beta, EcDHFR, MtFABG, retransformed failed DH5alpha plates, made miniprep cultures of GSK3 Beta, made 1L LB Media to be autoclaved manana, Journal Club/ Monday meeting

HSRA: made AMP, made 80% glycerol for glycerol stocks, FRI transformed Tem-1b BL21 and DH5a

Meeting Presenters:

FRI - Abigail, Nik ARI - Mai, Hannah HSRA - Jonathan, Harihar, Srinidhi, Akshitha

Tuesday - June 17th

2-3:50 - Middle schoolers visit - Mentors in Charge: (changed to Tuesdays this year from Wednesdays)

Everything is Tuberculosis

- John Green -

ALL

-

FRIers

- Target Disco
 - Talk about targets complete form
 - Targets page (VDS Portal)

Va Dal

- YpDalaDala
 - Yersinia pestis - peptidoglycan crosslinking cell membrane formation
- MtDalaDala
 - Mycobacterium tuberculosis peptidoglycan crosslinking cell membrane formation
- EcDHFR
 - E. coli precursors for DNA and amino acids
- PaDXR
 - Pseudomona aeruginosa bacterial Doozy

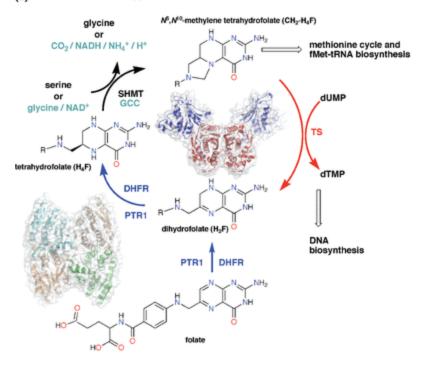
- https://www.uniprot.org/uniprotkb/Q9XFS9/entry
- 1-Deoxy-d-xylulose 5-phosphate reductoisomerase (DXR) is a key enzyme of the 2-C-methyl-d-erythritol 4-phosphate (MEP) pathway operating in several pathogens, including *Mycobacterium* and *Plasmodium*. Since a DXR homologue is not present in humans, it is an important antimicrobial target.

-

- YpFabG
 - Yersinia pestis- fatty acid biosynthiesis cell membrane formation
- MtFabG
 - Mycobacterium tuberculosis fatty acid biosynthiesis cell membrane formation
- MtInhA (fabl)
 - Mycobacterium tuberculosis- fatty acid biosynthiesis cell membrane formation
- LmPR -
 - Leishmania major protozoan parasite organism Pteridine reductase antifiolate - similar pathway to DHFR
 - https://pubs.acs.org/doi/10.1021/jm901059x

-

(a) Folate metabolism in trypanosomatids and contributions from PTR1 and DHFR-TS



(b) Biopterin reduction by PTR1

- GSK-3 beta
- Kinase involved in ALzheimers
 - Requires different purification

- Spin down o/n cultures
- VPipe PCR
 - Using primers we diluted last week
- Protocol Agarose Gel DSv3
- QIAprep® Spin Miniprep Kit
- Nanodrop

- Submit to Sequencing
- Large culture

-

- Transform YopH into BL21(DE3) someone XIYA AND BEDOUR *one 10ul one
 50ul KEERTANA TO MAKE PROTEIN
 - For expression for enzyme assay testing
- Everyone grow up some colonies of pNic-Bsa 4 in pairs to use for your PCR down the road - XIYA to make 8 x 4ml cultures
 - Will need to miniprep

-

- Purple Protein + Enzyme Project (PPEP) discuss w/ Dr. B
 - Goal: couple a purple protein 'tag' onto one of our target enzymes for easy identification during purification. This would be use in future HRI projects and potentially as a shared resources. (eventually want to do this in KPC-2 and TEM1b also)
 - Would this be a colorimetric indicator of expression levels?
 - Start Dh5alpha overnight cultures for pGBR22 and EhPTP for cloning project DONE
 - EhPTP
 - pGBR22 (Amp)
- EhPTP use Hubert and Quoc's primers Yes, do this since the control plasmid
- VR
- GBlock design session??

HSRA

- Sign in
- Talk about how much antibiotic needs to be pipetted onto each disc
- Transformation?
- Making LB media?
- Glycerol stocks day 2?

-

_

ARIers

- Liquid nitrogen training?
- Make SDS PAGE gels (make new Tris-HCl buffers both 6.9 and 8.8 pH)
 - ProtocolHomemadeSDSPageGelsVDS v6d

MENTORS:

- ARI: Post up signage (this week) -
- Make a laminated sign with pic and brief instructions on organizing -80degC freezer
 - Pic of 50 ml tube in a rack for Snap Frozen protein samples
 - Glycerol stocks go in boxes by semester (?) in cryovials
 - Except HRI glycerol stocks
 - Need pics of cyrovials and glycerol stock labeling and a box.

ENTER WHAT WAS COMPLETED TODAY (so we can plan for tomorrow).....-

- FRI: discussed target selections, saw which DH5a/BL21 plates failed again, made large cultures from small cultures, miniprepped various targets (stored... to be sent to sequencing tomorrow), autoclaved 1L LB Media, made miniprep cultures of pNIC Bsa4 and PfDXR, spun down GSK3-Beta (miniprep pellet), transformed YopH in BL21 and GSK3-Beta in DH5alpha,
- HSRA: made LB media, made Amp disc (25uL), day 1 of glycerol stocks, made small cultures
- ARI: homemade SDS PAGE gels, miniprep of DH5a overnight cultures, BL21 transformation

Wednesday - June 18th 2025

ALL

_

FRIers

- Pick up GBlocks in FNT

- Store in a new box labelled 2025 PIPE Cloning
- PCR VPipe
- Spin down expressions
- spin down cultures for mini -preps
- Start Dh5alpha overnight cultures for pGBR22 and EhPTP for cloning project GANESH
 to make 4x 4ml per plasmid
- Ideas
 - Make buffers for Homemade SDS-Page gels (the 2 Tris ones of different pH)
 - ProtocolHomemadeSDSPageGelsVDS
 - Tips for Optimal SDS-PAGE Separation | Rockland

HSRA

_

-

_

ARIers

_

Make SDS-PAGE Gels?

MENTORS:

_

- FRI (start this week) maybe Thursday since no HSRA
 - Make a 'map' for -80degC freezer and print for door.

ENTER WHAT WAS COMPLETED TODAY (so we can plan for tomorrow)....

- FRI: discussed protein targets vs DNA targets, transformed GSK3-b BL21 and YpDalaDala BL21, spun down large cultures, added lysis buffer/ sonicated cell pellets and stored in Julio (pellets not sonicated stored with lysis buffer), picked up g-blocks (stored in Julio), made miniprep cultures of GSK-3beta, pGBR-22, and EhPTP, made small cultures of YopH
- HSRA: made base broth, made large cultures, made 3 glycerol stocks of both Tem-1b and EhPTP
- ARI: completed BL21 transformation, additional miniprep, began purification

Thursday - June 19th 2025

ALL

_

FRIers

- Sonication -
 - (freeze immediately after if not going to purify today)
- Make new purification buffers (already done this summer?)
 - Wash, elution buffers
- DR B TO PRINT- Homemade SDS Page,

-

- PCR VPipe
- Start up the VR headsets and make sure they are logged into Nanome and are updated -

HSRA

Sign in on sheet

_

- HRI PROJECT

-

- Transformation re-do into BL21(DE3) for TEM1b and EhPTP (or move to MONDAY WEEK 3) - YES, move to week 3.

ARIers

- Sonicate expression samples
 - Store in -20 deg after sonication (wait on hi-speed spin)

_

-

MENTORS:

ENTER WHAT WAS COMPLETED TODAY (so we can plan for tomorrow)..... -

- FRI: continued sonicating, made 1L LB Media and 3L Base Broth to be autoclaved tomorrow, miniprepped various targets, sent various samples to DNA sequencing, attempted SDS page gel, miniprepped GSK3beta and EcDHFR, made large (hurried) cultures of YpDalaDala and GSK3beta, made large culture for YopH
- HSRA: I spundown large cultures, kept the pellets, transformed Tem-1b and EhPTP BL21
- ARI:

Friday - June 20st 2025

ALL

- Lab notebook checks
- Read Journal Club for next Week 4 (not next week) group meeting
 - https://sites.google.com/site/vdsstream/virtual-drug-screening-stream-a-k-a-virtual-cures/journalclub?authuser=0
 - Hsp90 Paper: Hsp90 paper

FRIers

- Lab notebook checks
- 1 or 2 on B's ???? (TBD)
 - Each VDSer pops into Dr. B's office for a quick checkin to see how your summer is going. Come solo or with a friend.

- Have we made purification buffers?

- HiSpeed spin for expression samples

- (if plan to purify, if not purifying today, then store sonicated sample in -20degC)
- Purification & Storage -
 - Should we try Liquid Nitrogen snap freeze?
 - If so, need to get LN2
- Prepare samples for SDS-Page (add blue juice and heat block them)
- Make TE Buffer
- PCR VPipe
- Have we made the 2 Tris buffers for Homemade gels?
- Make Homemade SDS-Page gels
 - ProtocolHomemadeSDSPageGelsVDS

SAVE FOR NEXT WEEK

- Characterization using Homemade SDS gels
 - ProtocolProteinCharacterization_ARI-FRI-VDS_v2c

HSRA

- Sign in on sheet
- Lab notebook checks
- _
- _

ARIers

- Lab notebook checks

MENTORS:

- Lab notebook checks
- _
- Get image of weekly sign in and sign out and send to Dr. B

ENTER WHAT WAS COMPLETED TODAY (so we can plan for tomorrow).....

- FRI: autoclaved LB Media and LB Broth, spun down large cultures from 6/19 and stored cell pellets, started purification (stored in column in Julio), made 6 SDS page gels, marked acrylamide for chemical waste
- HSRA: added Tris-NaCl, imidazole, and lysozyme to previously spundown samples, made agar plates (staff), made 500mL of LB media
- ARI:

Saturday

Sunday

DELIVERABLES BY END OF WEEK XXXXX

ALL

WEEK 3 - Jun-23

OVERALL ITEMS FOR WEEK 3 (by group)

ALL

Update Daily Conflict schedules on the Conflicts spreadsheet

ARIers

- Plasmid Stock Depositions, AddGene
- Site Directed Mutagenesis to make variant of Target
 - Primer design
 - Primer ordering

- SCM PCR w/ agarose gel pseudo- verification
- Target E, P, C
 - Express another batch of protein
- DSF
 - Next Steps: Ion Exchange
 - Next Steps: FPLC
 - Next Steps: Enzyme Assay test on EhPTP
 - Next Steps: Crystal Trays

FRIers

_

- Start VPipe PCRs all
 - Use existing pNIC-Bsa4 but don't use it all up
- Each person start o/n culture of pNIC-Bsa4 to use for their own miniprep of it to keep for VPipe PCR
- Target E, P, C
 - Next Steps: DSF
 - Next Steps: Enzyme Assay test on YopH
- Plasmid project work towards Plasmid Stock Deposition & AddGene
- Tail Primer Design protocol (computer based) for YpDFHR and YpFabG
 - We already have the primers, but do the design process to better understand what is going on
 - ProtocolPCR PrimerDesignTails for PIPE CloningVDS
 - Order tail primers for iPIPE (Dr. B)

HSRA

- Antibiotic disc testing
- Make glycerol stocks and plasmid stocks
- Express, purify, characterize proteins
- iCn3D
 - NEXT WEEK (S):
 - iCN3D con't...
 - VR Headsets
 - Continue protein work to DSF & Enzyme Assays

MENTORS:

-

DAY TO DAY ITEMS

Monday - DATE June 23th

ALL

- 1 pm (WCH 1.110) NOTE: ROOM CHANGE BACK TO ORIGINAL ROOM
 SHAREBACKS & Group meeting
 - Summ25 Shareback Slides
- -NO Journal Club in WEEK 3 (wait til Week 4)
 - Add final figures, add 2 reflections

FRIers

- Group meeting 1:30 pm
- Submit mini prep samples from last week to sequencing
- Nanodrop protein samples (for those who purified last week)
- Make and run agarose gel for checking mini prep
- Add link to your Research Page at the top of this doc
- DR. B to print- VPIPE

HSRA

- Group meeting 1:30 pm

ARIers

- Group meeting 1:30 pm
- Characterization?
- Run agarose gel of practice PCR (re-do)?

_

MENTORS:

ENTER WHAT WAS COMPLETED TODAY (so we can plan for tomorrow).

- FRI: Monday meeting, sharebacks, cont. purification, sonication, and preparing samples for DNA sequencing
- HSRA: sharebacks, made antibiotic testing plates, made more amp discs, made cef discs
- ARI:

Tuesday - June 24th

2-3:50 - Middle schoolers visit - Mentors in Charge: (changed to Tuesdays this year from Wednesdays)

ALL

- VDS Plasmid Archive (mentors)
 VDS Plasmid Stocks (Students)
 VDS Plasmid Vault (Dr B)
 - ARI-VDS Plasmid Student Archive

FRIers

Making SDS-PAGE gels (first attempt/second for some) ?

- VPIPE
- Cont. purification
- Submit to sequencing
 - Check sequencing for those who submitted

HSRA

_

ARIers

- SDM primer design

_

MENTORS:

ENTER WHAT WAS COMPLETED TODAY (so we can plan for tomorrow).....

- FRI: continued purification, some submitted to sequencing, some stored purification column, started VPIPE (pNIC), made small cultures for another expression round this week
- HSRA: purification (day 1 and 2)
- ARI:purified protein, purified DNA, continued protein expression, submitted some samples for sequencing, ran SDS-PAGE, DSF?

_

ARI-VDS

Wednesday - June 25th

ALL

_

_

FRIers

- Continue protein purification
- Grow up pNIC Bsa4
- Nanodrop elution samples
- Check DNA sequencing
 - On personal computer
- Continue with autoinduction
 - Large large cultures (1000mL each?)
 - Or making small cultures
- Research pages
- Agarose gel for VPIPE samples?

- DR B TO PRINT-- agarose gel prep

HSRA

ARIers

-

MENTORS:

- biowaste - submit a request for pickup?

-

ENTER WHAT WAS COMPLETED TODAY (so we can plan for tomorrow).....

FRI- continued purification, stored snap frozen elution samples in -80, spun down lysed, sonicated, and stored MtFabG, ran agarose gel (and made agarose gel) on VPIPE samples from 6/24, ran more VPIPE, continued autoinduction and made small cultures for cultures that weren't made yesterday, made 10 pNIC-Bsa4 cultures for miniprep, made autoinduction media and LB media to autoclave tomorrow

HSRA-

ARI -

Thursday - June 26th

ALL

-

.

FRIers

- Characterization?
 - DNA/Protein?
- Make more LB Media, Autoinduction Media, Sugar Mix?
- Primer design activity in computer lab?\
 - Welch 2.128 @1pm
 - YpDHFR and Pf6PGDH
- Cont. purification and autoinduction for protein targets

HSRA

-

ARIers

-

_

MENTORS:

ENTER WHAT WAS COMPLETED TODAY (so we can plan for tomorrow).....

FRI - autoclaved LB media and autoinduction media, spun down large cultures and stored cell pellets, continued with autoinduction, spun down mini prep and stored in Julio, made more LB media and autoinduction media to autoclave tomorrow, primer tail design activity in computer lab, started purification and stored column in 4deg, prepped sample for characterization, sonication of a pellet, stored VPIPE samples from 6/25

HSRA

ARI

-

Friday- June 27th

ALL

- Read Journal Club for next monday's group meeting -
 - for Journal Club
 - Journal Club Summer 2024
 - Hsp90 Paper: Hsp90 paper
- Lab notebook checks
- Update Research pages: <u>LinksToResearchPages_VDS</u>

FRIers

- Tail primer design session?
- Lysis buffer re up
- Autoclaved
- Purification cont.
- Characterization
 - For vpipe and protein?
- Sugar mix?
- Check DNA sequencing
- *journal club 2 and research pages updates
- Autoclave PCR tubes or eppendorfs?
- Update Research pages: LinksToResearchPages VDS

HSRA

ARIers

- Update Research pages: LinksToResearchPages VDS

MENTORS:

ENTER WHAT WAS COMPLETED TODAY (so we can plan for tomorrow).....

- FRI: autoclaved LB Media and Autoinduction Media, spun down large cultures (some stored w/ lysis buffer, some sonicated) and stored in Julio, miniprepped samples of pNIC Bsa4, SDS page gel for various protein samples, discussed research pages, shareback slides, journal club, and lab notebooks, purification and snap froze elution samples to store in -80
 - NEXT WEEK: need to make more lysis buffer... finish organizing -80
- HSRA:
- ARI:

Saturday

Sunday

DELIVERABLES BY END OF WEEK XXXXX

ALL

HSRA, FRI and ARI, off for July 4th (Friday) - what about Thursday? - Thursday is optional HSRA off for July 3rd (Thursday)

OVERALL ITEMS FOR WEEK 4 (by group)

ALL

- Group meeting w/ Sharebacks on Monday
- Journal Club #2 on Monday

ARIers

- Express + Purify EhPTP
- Plasmid Stock Depositions, AddGene
- Site Directed Mutagenesis to make variant of Target

- Finish up Primer Design and Ordering, if not done
- For Week 5: SCM PCR w/ agarose gel pseudo- verification
- Target E, P, C
 - Express another batch of protein
- DSF
- Prepare reagents for: Ion Exchange
 - Next Steps: FPLC
 - Next Steps: Enzyme Assay test on EhPTP
 - ProtocolEnzymeSpecificActivityVDSv1
 - ProtocolEnzymeTest Phosphatase VDS Fall13 varyEnzyme
 - Next Steps: Crystal Trays

FRIers

- Protein work
- Someone Express + Purify YopH
 - For eventual enzyme assay
- Plasmid stock depositions & AddGene deposits (<u>Beckham depositions</u>)
- Glycerol stocks of DH5alpha and BL21 cells for -80degC
- Cloning work
 - Someone make pNIC-Bsa4 for miniprep for everyone (already done?)
 - VPipe cloning

HSRA

- Antibiotic testing assays (disc diffusion)
 - Plan for what we want to do with next tests
 - What can we hypothesize
- DSF on your purified proteins
- Do the VR protocol for TEM1 ?

_

MENTORS:

_

DAY TO DAY ITEMS

Monday - DATE June 30th 2025

ALL

- 1 pm (WCH 1.110) SHAREBACKS & Journal Club
 - Summ25 Shareback Slides
- Journal Club

- JournalClubSumm25
- Hsp90 Paper: Hsp90 paper

FRIers

- Lysis buffer
- Plasmid stock depositions
 - DNA targets

-

HSRA

- Request 3D printing of TEM-1 from DIY MakerSpace (go over there and talk to them). ???
 - See file in <u>HRIWorkFlowCurriculum v.2</u>

ARIers

MENTORS:

ENTER WHAT WAS COMPLETED TODAY (so we can plan for tomorrow).....

- FRI: sonicated and stored cell pellet, made miniprep cultures, made lysis buffer, started purification, lysozyme DSF, small cultures, monday meeting, shareback slides, journal club #2, starting preparing column for GSK-3 beta purification,
 - PfFabG 50mL small culture
- HSRA:
- ARI:

Tuesday —-----July 1st 2025—-----

2-3:50 - Middle schoolers visit - Mentors in Charge: (changed to Tuesdays this year from Wednesdays)

ALL

- Check out PDB 101
 - Dr Olmos was a VDSers!
 - 1st author of the paper
 - https://pdb101.rcsb.org/motm/307
 - RCSB PDB 6B68: Beta-Lactamase, 100ms timepoint, mixed, shards crystal form

FRIers

-

ARIers

MENTORS:

ENTER WHAT WAS COMPLETED TODAY (so we can plan for tomorrow).....

FRI: sonicated and stored cell pellets, spun down large cultures and stored, made large cultures from small cultures, made mini prep cultures for remaining DNA plasmids, purified and snap froze elution samples

HSRA

ARI

Wednesday -----July 2nd 2025------

ALL

_

FRIers

- V PIPE?
 - Characterization of last week's VPIPE on agarose gel
 - Protein purification
 - Continue autoinduction
 - Miniprep?
 - And submitting to sequencing
 - Homemade SDS page gels?
 - YopH purification (maybe another round of expression?)
 - To be used for enzyme assay

HSRA

-

VR Headsets ? or save til last week after poster is submitted?

ARIers

-

-

MENTORS:

ENTER WHAT WAS COMPLETED TODAY (so we can plan for tomorrow).....

FRI: spun down large cultures and stored, protein purification and storage of elution buffers, miniprep, made sds page gels, made agarose gel and ran vpipe samples and stored in 4deg, made another agarose gel stored in 4deg

HSRA ARI

Thursday -----July 3th 2025—-----

ALL

- Optional day for FRI and ARI
- No HSRA today

FRIers

- Solutions?
 - Lb media, autoinduction media, sugar mix, protein purification solutions
- autoclave solids?
 - Pcr tubes, eppendorf, collie rollers?
- VPIPE

ENTER WHAT WAS COMPLETED TODAY (so we can plan for tomorrow).....

- FRI: VPIPE, characterization of protein samples, miniprep, sonication, protein purification, made various solutions to autoclave next week (stored in bottles)
- ARI:

Friday -----July 4th 2025—-----

ALL

- NO LAB TODAY 🎇 🎆 🚞

Saturday

Sunday

DELIVERABLES BY END OF WEEK 4

ALL

WEEK 5 - Jul-7 Monday +++++++++

OVERALL ITEMS FOR WEEK 5 (by group)

ALL

- Science documentary? maybe next week on Tuesday
- Group meeting & Sharebacks on Monday
- Cool MtInhA (fabl) papers:
 https://onlinelibrary.wiley.com/doi/full/10.1111/cbdd.70152?campaign=woletoc

ARIers

- Finish Primer Design and Put Primers on the WishList in IDT for Dr. B to order
- DSF's
- Variant PCRs SDM
- Crystal trays of Target protein?
- Ion Exchange
- Make EhPTP for Enzyme Assays

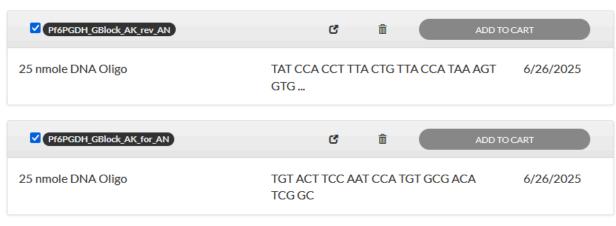
FRIers

- iPipe Primers are ordered. Once they come in, dilute them. And store in shared PIPE cloning box

Saved Items



.



- Express more protein & save pellets

DSF - in groups w/ lysozyme

- Make fresh lysozyme
- Determine melting temp
- Achieve precise curves
- Then do with Target protein
- Test one inhibitor (save for once you get precise curves on your Target)
- Plasmid Stock Depositions
 - 2 for Plasmid Stocks
 - 1 tube for Dr. B's Plasmid Vault (with unlabelled circle on top, but with side labelled)
- PIPE cloning
- Make YopH for Enzyme Assays
- Enzyme assays

HSRA

- Start prepping posters
- HRI module
 - Antibiotic disc testings
 - Amp, Tazo, Amp+Tazo (repeat on 2 separate days)
 - Cef, Taco, Cef+Tazo
 - SOSC
 - Antibiotic disc testing with IPTG (repeat on 2 separate days)
 - Characterization on proteins?
 - DSF on proteins
 - Enzyme assay on EhPTP ? pNPP repeat?
 - Enyzme assay on TEM-1b and KPC-2 ? nitrocefin
 - Increasing amounts of protein, increasing amounts of Nitrocefin
 - Time based measurement

MENTORS:

-

DAY TO DAY ITEMS

ALL

- 1:30 pm (WCH 1.110) SHAREBACKS
 - Summ25 Shareback Slides
- -NO Journal Club in WEEK 5 (wait til Week 6)

ARIers

_

Repeat DSFs until get precise curves

FRIers

_

HSRA

_

MENTORS:

ENTER WHAT WAS COMPLETED TODAY (so we can plan for tomorrow).....

- FRI: submit to DNA sequencing, gel staining/destaining/drying, solution making, autoclaved LB Media and Autoinduction media, monday meeting/ sharebacks, start of protein purification of GSK 3beta
- _
- HSRA
- -
- ARI

_

Tuesday July 8th, 2025

2-3:50 - Middle schoolers visit - Mentors in Charge: (changed to Tuesdays this year from Wednesdays)

ALL

_

ARIers

- Finish Primer Design and Put Primers on the WishList in IDT for Dr. B to order
- Ion Exchange

FRIers

-

- etc

HSRA

_

MENTORS:

ENTER WHAT WAS COMPLETED TODAY (so we can plan for tomorrow).....

- FRI: DSF lysozyme calculations, protein purification, SDS Page Gel characterization,
- HSRA
- ARI

ALL

_

FRIers

- UPDATE your Research Pages!!
 - Important to put your data and results images and numbers here
 - Include the specific name of the protocol you used -but don't have to provide the link - unless useful to you
 - We will plan on research page checks on Thursday
- DSF lysozyme
- Transformation- pNicBsa4 Dh5a and MtInhA-pNicBsa4 Dh5a
- SDS Page Gels
- Characterization
- VPIPE
- Plasmid stock deposition

HSRA

-

ARIers

_

MENTORS:

ENTER WHAT WAS COMPLETED TODAY (so we can plan for tomorrow).....

- FRI: DSF lysozyme, prepared nitrocefin working stock (stored in samtonia box in Julio) and buffer for enzyme assay, protein purification, transformation for DH5a, submitting to DNA sequencing
- HSRA
- ARI

Thursday —---July 10th, 2025------

ALL

-

FRIers

- Enzyme assay... cont. to Fri?
- Check DSF results

HSRA

-

ARIers

_

MENTORS:

- Check VDSers Research pages
 - Need 'pieces of data' and brief analysis for the weeks

ENTER WHAT WAS COMPLETED TODAY (so we can plan for tomorrow).....

- FRI: Checked DSF results, started enzyme assay w/ YopH, protein purification (GSK3Beta), SDS Page Gels, protein characterization, made autoinduction media to autoclave tomorrow, made sugar mix, submitting to DNA sequencing, picked up primers from FNT, VPIPE
- HSRA
- ARI

Friday —-----July 11th, 2025-----++++++++++LINK TO TOP

ALL

-

_

- Read Journal Club for next week
 - **JC3** WEEK 6
 - Journal Club Summer 2025

- Prasannan, P.; Suliman, H. S.; Robertus, J. D., Kinetic analysis of site-directed mutants of methionine synthase from Candida albicans. *Biochem Biophys Res Commun* **2009**, *382* (4), 730-4.
- <u>PrasannanPriya MetSynthaseKinetic RobertusBiochemBiophysResComm2009.</u> pdf

-

FRIers

- Autoclave solutions
- VPIPE characterization
- Enzyme assay YopH cont. (in pairs)
 - Get spectrophotometers warmed up
- Mini prep/ submit to DNA sequence
- SDS Page Gels?
- Protein purification
- Destaining of SDS Page Gels
- IPIPE Primer Dilution

HSRA

- Posters due to HSRA. Sam or Dr. B to submit
 - Submit by 12 pm (noon)

ARIers

-

MENTORS:

-

-

ENTER WHAT WAS COMPLETED TODAY (so we can plan for tomorrow).....

- FRI: VPIPE characterization, protein characterization, YopH purification, cont., YopH enzyme assay, destaining and drying SDS Page Gels, IPIPE primer dilutions, miniprepped, autoclaved autoinduction media
- HSRA:
- ARI:

Saturday

Sunday

DELIVERABLES BY END OF WEEK 5

ALL

WEEK 6 - Jul-14 Monday, 2025

OVERALL ITEMS FOR WEEK 6 (by group)

ALL

- Update Daily Conflict schedules on the Conflicts spreadsheet
- Update weekly/monthly conflicts on Landing Page as a comment
 - - Journal Club #3
 - <u>JournalClubSumm2</u>5
 - Prasannan, P.; Suliman, H. S.; Robertus, J. D., Kinetic analysis of site-directed mutants of methionine synthase from Candida albicans. *Biochem Biophys Res Commun* 2009, 382 (4), 730-4.
 - PrasannanPriya MetSynthaseKinetic RobertusBiochemBiophysResComm2009.
 pdf
- Documentary Viewing Tuesday at 3:30 in NMS 1.120 (door code: 4882)
- Bring snacks/drinks if you'd like

ARIers

-

FRIers

_

HSRA

WED: 1pm - HSRA poster sessions
 Welch Grand Hallway

•

MENTORS:

-

DAY TO DAY ITEMS

Monday - DATE July 14th, 2025-----+++++++++LINK TO TOP

ALL

- 1:30 pm (WCH 1.110) SHAREBACKS & Journal Club
 - Summ25 Shareback Slides
- JC3 WEEK 6
- Journal Club Summer 2025
- Prasannan, P.; Suliman, H. S.; Robertus, J. D., Kinetic analysis of site-directed mutants of methionine synthase from Candida albicans. *Biochem Biophys Res Commun* 2009, 382 (4), 730-4.
- <u>PrasannanPriya_MetSynthaseKinetic_RobertusBiochemBiophysResComm2009.pdf</u>

_

FRIers

- MONDAY meeting, journal club #3, shareback slides
- SDS page gels?
 - Need to increase stock of homemade gels
 - People who haven't made them yet should make some
 - Good to try to make two in case one fails
- IPIPE?
 - Primers stock solution and working dilutions in Julio (IPIPE box)
- Rerun VPIPE?
- Plasmid stock deposition?
 - How have sequencing results come back?
- Mini prep/ midi prep?
 - Should try midi prep cultures if already tried miniprep at least 2x
- Small cultures autoinduction
- VR Goggles

HSRA

_

- Poster preparation time

ARIers

MENTORS:

-

ENTER WHAT WAS COMPLETED TODAY (so we can plan for tomorrow).....

- FRI
- HSRA
- ARI

Tuesday—-----July 15th, 2025-----

No Middle schoolers this week or next

ALL

.

Documentary Viewing (Tuesday) @ 3:30 PM

- In NMS 1.120

-

FRIers

_

HSRA

-

ARIers

-

MENTORS:

_

ENTER WHAT WAS COMPLETED TODAY (so we can plan for tomorrow).....

FRI

HSRA

ARI

`Wednesday—-----July 16th, 2025-----++++++++++LINK TO TOP

ALL

-

-

FRIers

- Visit HSRA poster session
- DSF lysozyme again
- YopH enzyme assay (phosphatase)?

- Continuous B Lactamase enzyme assay?

HSRA

- Poster Presentation WELCH GRAND HALLWAY
 - Session A: 11 am 12 pm: other streams
 - Session B: 1 pm 2 pm: others streams + VDSers
- Last Day 😥

ARIers

- Visit HSRA poster session
- EhPTP enzyme assay?

MENTORS:

ENTER WHAT WAS COMPLETED TODAY (so we can plan for tomorrow)...... FRI

ARI

Thursday------July 17th, 2025------

ALL



-

FRIers

- PIPE clonign
- _

ARIers

- Continuous B Lactamase enzyme assay?

MENTORS:

ENTER WHAT WAS COMPLETED TODAY (so we can plan for tomorrow).....

- FRI
- ARI

Friday —-----July 18th , 2025----++++++++<u>LINK TO TOP</u>

ALL

- Lab notebook checks
- -

FRIers

- YopH enzyme assay?

- Continuous B Lactamase enzyme assay?

AR	lers

MENTORS:

ENTER WHAT WAS COMPLETED TODAY (so we can plan for tomorrow).....

- FRI:
- _
- ARI:

Saturday

Sunday

DELIVERABLES BY END OF WEEK 6

ALL